Gene Pulser® Electroprotocols

* We recommend adapting this protocol to use the Gene Pulser electroporation buffer (catalog #165-2676, 165-2677), which increases cell viability and transfection efficiency in mammalian cell lines.

Cell Type	Mammalian, adherent, suspension,
Species Used	Monkey, Vero, kidney cells; Human, C-4I, cervical carcinoma cells
Molecules Electroporated	DNA: circular, 9.2kB, expression vectors

Before the Pulse

Cell growth medium	M199 (GIBCO/BRL, Sigma)
Wash solution	M199
Growth phase at harvest	3 x 10 (6) cells / ml
Pre-pulse incubation	None

The Pulse

Electroporation Temperature	Room temperature
Electroporation Medium*	M199
Cell Density	3 x 10 (6) cells / ml
Volume of Cells	0.3 ml / pulse
DNA Concentration	100 μg DNA / pulse
DNA Resuspension Buffer	distilled water
Volume of DNA	5 μl
Instruments Used	Gene Pulser® apparatus & Capacitance Extender
Cuvette Gap	0.4cm
Voltage	0.250 kV
Field Strength	0.625 kV/cm
Capacitor	500 μF
Resistor	(Pulse Controller) Ω none
Time Constant	Not given

After the Pulse

Outgrowth Medium	M199 - DMEM /F12,5% FBS,5% serum supplement, 1% pen/ strep, 400 μg G418
Outgrowth Temperature	37°C
Length of Incubation	24 hours recovery before selection
Selection Method or Assay Used	G418
Electroporation Efficiency	1.3 / μg DNA
Per Cent Survival	Not given

Relevant Publications and/or Comments

Note: exponential values designated in parentheses.

Name of Submittor

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Date Submitted 8/24/91

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